## STATUS OF THE AIRLIE ISLAND CTENOTUS, CTENOTUS ANGUSTICEPS (LACERTILIA: SCINCIDAE), WITH NOTES ON DISTRIBUTION, HABITAT AND GENETIC VARIATION

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## ABSTRACT

The scincid lizard, *Ctenotus angusticeps* was previously only known from a single island (Airlie Island) and two disjunct mainland localities in Western Australia. As a result of its restricted distribution and little documentation, it has for some time been listed under both state and federal legislation. Recent surveys have revealed additional populations along the northwestern coast, which appear to be highly restricted to specific habitats, primarily salt marsh communities adjacent to mangroves where lizards shelter down crab holes. Genetic analysis of mitochondrial DNA shows very little variation between populations, suggesting a single widespread population or recent radiation. In light of this additional information the conservation status of *C. angusticeps* is evaluated.

## INTRODUCTION

The scincid genus *Ctenotus* is Australia's most diverse with currently 103 described species (Wilson and Swan 2013) displaying frequent sympatry and morphological similarity. As a result, numerous studies on habitat separation and ecological interactions have been carried out (e.g. Read 1998; James 1991; Sadlier 1987; Pianka 1986). However, for many species, published ecological data is minimal with their species descriptions representing the only source of available information.

Ctenotus angusticeps was first described by Storr (1988) from a series collected during a biological survey on Airlie Island, 35km NNE of Onslow on the northwestern coast of Western Australia (Figure 1). Presumably due to its restricted distribution on a small island (0.28km<sup>2</sup>) and the comments made by Browne-Cooper and Maryan (1990)



**Figure 1.** Updated distribution map for *Ctenotus angusticeps* showing a very discontinuous range along the northwestern coast of Western Australia due to an association with a very specific habitat. Abbreviations: Ck: Cape Keraudren, Ps: Port Smith, Th: Thangoo, Cc: Crab Creek, Wc: Willie Creek.

regarding habitat disturbances at the island, this species was listed as 'Threatened' in Western Australia [under the Wildlife Conservation Act Specially Protected Fauna Notice in 1990]. Since this listing, C. angusticeps has been found on the mainland at two very disjunct coastal localities, one on Thangoo Station, at the southern end of Roebuck Bay in the Kimberley region (Sadlier 1993) and the other at Port Hedland in the Pilbara region (Turpin and Ford 2011). Both mainland accounts provide information on ecology and habitat and make morphological comparisons between known populations. Currently, C. angusticeps has both a state and federal listing as Schedule 1 under the Wildlife Conservation Act 1950 and Vulnerable under the Environment Protection and Biodiversity Act 1999. Two other restricted range Ctenotus species C. lancelini and C. zastictus have the same listings. Due to its perceived vulnerable

Due to its perceived vulnerable status, one of the stipulated research priorities for C. *angusticeps* is to undertake survey work to locate additional populations in the intervening areas and determine the dis-

tribution of C. angusticeps along the northwestern coast of Western Australia (Threatened Species Scientific Committee 2008). Considering the distance between the two mainland localities and the lack of survey effort in its documented habitat thus far by experienced field zoologists, it seemed highly likely that additional populations would be located. Due to recent finding of C. angusticeps at Port Hedland (Turpin and Ford 2011), an area undergoing rapid port and/or infrastructure development by mining companies, a reconnaissance survey was conducted in December 2011 to determine its local distribution. relative abundance and habitat preference. This was followed by a broader-scale survey in May 2012, covering much larger areas to the north and south of Port Hedland to search for additional populations. Here we report on results of a targeted survey from Onslow to Broome in May 2012, presenting new location records, molecular examination of known populations and further notes on habitat preferences and other observations. We also provide a suggestion of a common name change and a discussion relating to its current conservation status.

### METHODS

Experienced field zoologists familiar with the identification of *Ctenotus* spp. and with considerable survey experience conducted daytime searches for C. angusticeps from 2–10 May 2012. This involved two teams of three people investigating areas of potential suitable habitat as described by Sadlier (1993) and Turpin and Ford (2011), and the immediate surrounding habitats between Onslow and Broome. Potential habitat was initially examined using vegetation maps, aerial imagery and Google Earth imagery, and permission was sought from landowners to conduct any surveys on private land.

Searching methods involved slowly walking through areas considered suitable and visually observing active lizards until a positive identification was made. Once confirmed, attempts to capture some individuals were made by hand or by using a circular metal ring to entrap a lizard within low vegetation. Due to the extensive survey area to be assessed and time restraints, the use of more permanent trapping techniques was not feasible.

Photographic records were taken of the habitats where C. angusticeps were found along with images of individual lizards from most localities. Tissue samples (tail-tips) were taken from ten C. angusticeps captured, and representative voucher specimens were also collected and lodged with the Western Australian Museum (Tissues R634-43; Specimens R172522, R172876-78). Each lizard captured was then weighed. measured and released at capture site. The retention of tissue and specimens was done in accordance with Regulation 17 licence to take fauna for scientific purposes (Number SF008623) issued by the Department of Environment and Conservation, Western Australia.

We analysed 580 base pairs of mitochondrial DNA for the ND2 gene from thirteen individuals, the rate of change in this mitochondrial locus has been estimated to be c. 1.3-1.4% Mvr in other skink species (Macey et al. 1997; Greaves et al. 2008). DNA was extracted using DNeasy tissue extraction kits (Qiagen) and the primers ND2 L4437 (Macey et al. 1997) and ND2r102 (Sadlier et al. 2004) were used. Standard PCR thermocycling protocols were used and amplicons were purified using either ExoSap-IT (USB Corporation, OH, USA) or QiaQuick PCR clean up (Oiagen). Purified templates were sequenced in both directions by Macrogen (Seoul, Korea). Sequences were edited and aligned with Sequencher v. 4.10 (Gene Codes, Ann Arbor, MI) and Geneious v5.5.6. Pairwise sequence divergence was calculated using MEGA 5. JModel test (Posada 2008) favoured an GTR+G model for the ND2 mtDNA data and accordingly MrBayes (Huelsenbeck and Ronquist 2001) was used to generate a maximum a postori tree (2.2 million iterations with 10% burnin) where parameters were surveyed (through ESS values) to ensure appropriate mixing. A sequence of Ctenotus robustus, the closest Ctenotus for which ND2 has been sequenced. was downloaded from GenBank (AY662548) and incorporated into the alignment as an outgroup.

## NEW LOCATION RECORDS

In the Port Hedland area, C. angusticeps was recorded at multiple localities, including

able 1. Summary of new	location records for	C. angusticeps recorded	in May 2012.
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LOCATION	DATE
Pretty Pool, Port Hedland	02/05/2012
Wedgefield, Port Hedland	02/05/2012
Redbank, Port Hedland	03/05/2012
Finucane Island, Port Hedland	03/05/2012
Boodarie Station, 12 and 16km W Port Hedland	03/05/2012
Lulu Creek, 7km E Karratha	06/05/2012
Beebingarra Creek, 15km E Port Hedland	03/05/2012
Cape Keraudren, 9km N Pardoo Roadhouse	04/05/2012
Port Smith, 25km NE Bidyadanga Community	07/05/2012
Thangoo Station, 28km SSE Broome	08/05/2012
Crab Creek, 16km ESE Broome	08/05/2012
Willie Creek, 25km N Broome	08/05/2012

Redbank (Turpin and Ford 2011), Finucane Island, Pretty Pool and Wedgefield. The species was also recorded immediately west of Port Hedland on Boodarie Station and east at Beebingarra Creek. We also recorded C. *angusticeps* from near Karratha, Cape Keraudren, Port Smith, Thangoo Station (Sadlier 1993) and both Crab and Willie Creeks in the Broome area (Table 1, Figure 1).

#### SEQUENCE DIVERGENCE

Intraspecific variation in the ND2 gene fragment for C.

angusticeps sampled from sites throughout their known range (Figure 2) up to 850 km apart was small; the thirteen sequences were 99.3% identical across the 580 nucleotide positions. The pairwise distance (using multiple models) in the ND2 gene was uniformly low  $\sim 0.6\%$ . While the Airlie Island population share a haplotype (based on a single transversion). the clade is embedded within the mainland diversity. The ND2 gene region used here has been investigated in other skink species where clades within the same species were much greater than observed



**Figure 2.** Maximum *a postori* tree of *Ctenotus* ND2 sequence. The tree was constructed in MrBayes (Huelsenbeck and Ronquist 2001) using thirteen *C. angusticeps* sequences (580 bp) along with *C. robustus* (AY662548) as the nominated outgroup. Posterior probabilities are shown on the nodes.

here (Greaves *et al.* 2008). Taken together the data suggest the sampled populations are conspecific. While the mtDNA ND2 genetic distance is very shallow it is acknowledged that to more accurately map the extent of gene flow between populations that polymorphic markers (such as STR's) may need to be evaluated. ND2 sequences have been deposited on GenBank with accession numbers (KF537351– KF537363).

## HABITAT AND ENVIRONMENT

Ctenotus angusticeps has been previously recorded as occurring in most habitat types on Airlie Island, such as tussock grasslands and Acacia coriacea shrubland with coastal Spinifex (Browne-Cooper and Maryan 1990), however this differs markedly to the vegetation occupied on the mainland at Roebuck Bav (Sadlier 1993). Turpin and Ford (2011) provided a detailed description of the habitat where C. angusticeps was recorded at Port Hedland that is consistent with the 'samphire shrubland along a mangrove margin' described by Sadlier (1993). Our survey effort was concentrated in these 'salt marsh communities' on the landward fringe of mangroves, which are numerous and patchy along the northwestern coast sometimes forming large vegecommunities (Van tation Vreeswyk et al. 2004). Although the habitats on Airlie Island and the mainland are markedly different, the primarily coastal distribution of C. angusticeps may suggest that the 'salt marsh communities' are the ancestral habitat of this species, with arguably the island population secondarily adapting to the habitat now present on the island. This fragmented distribution is mirrored in other fauna such as the legless lizard Delma borea and the murid rodent Pseudomys nanus, which also have relictual populations on islands off the Pilbara coast (Maryan *et al.* 2007).

Our observations confirm that C. angusticeps is very habitat specific, occupying areas broadly described as the landward fringe of salt marsh communities, vegetated with samphire and marine immediately couch grass adjacent to mangroves or some distance away, though still within close association. The dominant vegetation being the succulent samphires Tecticornia halocnemoides subsp. tenuis and Suaeda arbusculoides on clavev soils with a mixed herb and grass cover of Muellerolimon salicorniaceum and Sporobolus virginicus on sandy soils (Beard 1990). Ctenotus angusticeps was

**Figure 3.** The habitat at some of the locations where *C. angusticeps* were recorded showing the subtle differences in structure of samphire and marine couch grass on a sandy clay substrate with numerous crab holes. All locations become inundated during very high tides, (A) Karratha, (B) Cape Keraudren, (C) Crab Creek.



observed at some localities within the mangrove-lined creek edges but displayed a marked preference for the low open shrubland area that is subject to tidal influences with numerous crab holes on a heavy, greyish sandy clay substrate. At some locations where low sandy rises persisted with dense vegetation, especially marine couch grass, C. angusticeps was more numerous. These areas either formed a raised 'island' surrounded by bare. sparsely vegetated mud flats or a narrow strip (< 10m) growing through the samphire. Overall, the general structure of the samphire did vary subtly from very low, dense clumps interspersed with marine couch grass to very flat, prostrate clumps or medium-high shrubs surrounded by bare ground (Figures 3A–C).

Despite the apparent suitability of the habitat, C. angusticeps was not observed in the marginal. sandier Triodia-dominated vegetation, where other species such as C. saxatilis and C. pantherinus were prevalent. The occasional C. saxatilis was observed in the same habitat as C. angusticeps, particularly in the Port Hedland area, though in fewer numbers. An apparent barrier may be the Eighty Mile Beach area (Figure 1). where no C. angusticeps were detected, despite the presence of flat coastal plains of clayey sand with some samphire and marine couch grass; however there were no substantial mangroves with associated crab holes. We also did not record any C. angusticeps at

Derby, where ample mangroves and tidal creeks margin the King Sound. However from our observations, the extreme landward fringes of salt marsh communities were devoid of crab holes and appeared unsuitable for this species. We did however; observe the abundant species C. *inornatus* in these areas.

The salt marsh communities inhabited by C. angusticeps are associated with the more extensive mangrove communities, which are typically coastal plants subject to periodic tidal inundation and exhibit a number of anatomical adaptations and physiological characteristics which enable them to flourish under these conditions (Semeniuk et al. 1978). Not only is this saline environment harsh, subjected to extreme heat, wind, glare and tidal influences, it is also dynamic by virtue of the ever encroaching movement and recruitment of mangroves into new adjacent areas that are dependant on water levels and the effects of erosion. The mangroves are interconnected with the landward salt marsh communities by trapping sediment and creating areas of sand deposition for samphire and other species to grow. Therefore, it is feasible to assume that the environment for C. angusticeps would expand and contract over relatively short periods of time connecting and isolating lizard populations on a regular basis and correspondingly, if C. angusticeps were reliant on crab holes (see notes below) for shelter, then the creation of these holes would fluctuate seasonally and possibly change from year to year.

## USAGE OF CRAB HOLES

At all locations where С. angusticeps was detected, crab holes of varying sizes were present and as reported by Turpin and Ford (2011), these were often used by skinks to take shelter, evading our attempts to capture them. Initially, when adult lizards were observed they took shelter at the base of low vegetation, followed by retreating down a hidden hole among the samphire and marine couch grass. By contrast, juvenile lizards usually relied on the protective cover of the vegetation to avoid capture, often taking refuge amongst the dense tussocks of marine couch grass. They would do this with some ease and after being pursued, later being located deep within the tussock. In addition, both adults and juveniles were sometimes observed to retreat up into samphire vegetation, sometimes up to 30cm above the ground. Thus, this species seems to be opportunistic in selecting retreat sites upon pursuing, and the tendency and ability of lizards to utilise a range of refuge types, both above and below ground level, may be of great advantage in a relatively open environment where vulnerability to predation could be high.

To test whether lizards would utilise the crab holes even in the absence of perceived threat of being captured by us, we used a non-toxic fluorescent powder (Radiant Colour Series T1. DayGlo Colour Corporation, Ohio) to track their movements. Two adult C. angusticets at Redbank near Port Hedland caught early in the day were dipped in the 'powder' and released shortly afterwards (methodology adopted from Stark and Fox 2000; Bernd and Henle 2004). The next morning we tracked their movements from the previous day to separate crab holes where lizards sheltered overnight, showing that the lizards are using crab holes by choice.

In highly exposed and harsh environments, burrows and holes created by other animals would provide effective protection to lizards (Davidson et al. 2008). Many other species of Ctenotus shelter down holes, mainly excavated by insects, or by themselves (Taylor 1984) especially in areas where the abundance of cover in the form of rocks, logs, leaflitter, various hummock grasslands and deep earth cracks is less (B. Maryan and R. Lloyd pers. obs.). Similarly, there is minimal effective shelter where the C. angusticeps live apart from the crab holes and limited dense vegetation, thus the former could be an essential criterion in their habitat selection. In areas where there was artificial shelter (e.g. pieces of driftwood), some skinks took shelter beneath these, but they eventually disappeared down a crab hole.

Another interesting observation made regarding the use of crab holes was at Port Smith. where a lizard was observed entering a low, prostrate samphire on an incoming tide that soon became submerged without the lizard being re-sighted. After the tide receded, we continued our searches on the recently inundated ground that was very damp underfoot and sighted several active C. angusticeps. We assumed at the time, that the lizards were down crab holes, possibly utilising air pockets to sustain their residence, however this requires further investigation. The use of crab holes by reptiles in areas subjected to periodic inundation has been documented previously for other species of Australian reptiles occurring in tidal/ mangrove communities (e.g. Fordonia leucobalia. Myron richardsonii: Gow 1989: Cox 1991: Karns et al. 2005; Murphy 2007; R. obs.). In Somaweera pers. southwestern Western Australia, the skink Lissolepis luctuosa and snake Elapognathus minor, species that do inhabit seasonally inundated wetlands, have been found in completely submerged freshwater crayfish holes (B. Maryan pers. obs.). Burrows of other crustaceans in mangrove communities are also used by an array of invertebrates (Ng and Kang 1988), fish (Al-Behbehani and Ebrahim 2010; Berti *et al.* 2008), frogs and other lizards elsewhere (Ng and Sivasothi 1999).

The apparent high frequency of crab hole usage by lizards in the salt marsh communities lead us to believe that these were an essential requirement for this species. This is reinforced further bv not observing anv C. angusticeps in areas without crab holes. Mangrove crabs could be considered as 'ecosystem engineers', a group of animals that physically create, maintain and modify their environment (lones et al. 1994). Hence the spatial distribution and abundance of species that utilise these holes could vary with the distribution of the particular species of crabs. The crab species inhabiting the northwestern coastline of Western Australia are diverse with many specific to mangroves and associated salt marsh communities (Jones and Morgan 1994). The most likely species digging the holes, where we observed C. angusticeps are the Fiddler Uca spp. and Bubbler Crabs Scopimera spp. Further studies are required to investigate whether the distribution of C. angusticeps is correlated to distribution of any particular type of crab as shown for some other species (e.g. Subramanian 1984: Berti et al. 2008).

### ACTIVITY

Ctenotus species are typically diurnal, but some display

nocturnal activity and are generally notoriously difficult to observe due to their cryptic nature. They maintain high average active body temperatures of 30-38°C (Greer 1989), and because the air and ground temperatures are timedependent, the temporal activities of different species must be 'geared' closelv to their thermoregulatory requirements. Ctenotus angusticeps inhabits an environment subjected to very high daytime temperatures with strong solar radiation. Observations made during our survey show that C. angusticeps can tolerate very high ground temperatures and thus be active throughout the day, moving at great speed from one clump of vegetation to the next. Even though May is considered to be a cooler time of the year along the northwestern coastline of Western Australia, we still recorded ground temperatures of 41-45°C when C. angusticeps was actively foraging. Due to the scope and size of the survey area between and Broome, areas Onslow considered suitable were surveyed at different times of the day (extended into the twilight hours), and individuals were still observed. However it is likely that more activity is concentrated in the early mornings and late afternoons (Browne-Cooper and Maryan 1990; Sadlier 1993), in accordance with the bimodal daily activity pattern shown by many lizards during warmer times.

The long diurnal period of activity observed by us could be adaptation to increased an foraging efficiency in a highly fluctuating environment. We observed individuals foraging both on the raised sandy patches and among the pneumatophores of mangrove plants during low tide. While no records of their diet exist, it is likely that they are opportunistic feeders, like many other species in the genus (Twigg et al. 1996; Read 1998). Future studies could investigate how they manage the potently high salt levels in their bodies, as arguably a component of their diet may comprise intertidal invertebrates.

### COLOUR AND PATTERN

The description of C. angusticeps by Storr (1988) was based on individuals collected from Airlie Island. The dorsal and lateral surfaces were described as dark olive grey with a mottled pattern consisting of black and whitish markings. These markings tended to be arranged on the side of the body in three series of 'ocelli'. e.g. short, whitish longitudinally elongate bars laterally edged with black, and the dorsal pattern being vague, but with some indication of a black vertebral stripe. Although Storr (1988) stated that C. angusticeps had a pattern of 'ocelli' on the lateral surfaces, it is more obscure and irregular when compared to C. pantherinus, which has a clearly-defined series of longi-



tudinal, dark-edged white spots or bars (see Plate 6, Storr *et al.* 1999). In general appearances, the pattern of *C. angusticeps* is more similar to *C. grandis*, a widespread arid-adapted species also flecked with white on the lateral surfaces.

Based on the description by Storr, comments by Sadlier (1993) and Turpin and Ford (2011), and our recent observations, it seems that C. angusticeps has consistent colouration and pattern morphology. This can be described as a faintly mottled pattern, particularly in adults, of short whitish to cream flecks or bars intermixed with blackish markings that tend to align longitudinally on the dorsal surfaces to form a vertebral stripe often extending along the tail, on an olive-grey or greyish to light brown ground colour (Figure 4). In some adult individuals at Karratha and Cape Keraudren, the lateral pattern is very weak with a strong indication of a vertebral stripe (Figures 4A-B), while others at Crab Creek had a bolder pattern (Figure 4C) similar to the juvenile colouration described by Turpin and Ford (2011). At all localities where C. angusticeps was observed, this colouration afforded them excellent camouflage on the sandy clay substrate when lizards 'froze' amongst the vegetation.

#### NEW COMMON NAME SUGGESTION

Given that C. angusticeps is now known from a much broader (albeit patchy) distribution on the coastal mainland in the Kimberlev and Pilbara biogeographic regions and is a species that occurs in a very specific habitat different from most other Ctenotus spp. in Western Australia (Storr et al. 1999), we recommend changing the common name 'Airlie Island Ctenotus'. The continued use of this common name incorrectly implies that C. angusticeps only occurs on this island. Based on available knowledge and as proposed by Wilson and Swan (2013) the 'Northwestern Coastal Ctenotus' is an appropriate replacement name.

# FUTURE AND CONSERVATION

The information collected on C. angusticeps during our survey has considerably added to the knowledge of this species. Prior to our surveys, the mainland distribution of C. angusticeps was only known from two very disjunct localities that were serendipitously made in а habitat that has received very little biological attention. This is underlined especially in the Pilbara, which is one of the most

<sup>◄</sup> Figure 4. Representative Ctenotus angusticeps from three localities along the northwestern coast of Western Australia showing the subtle colour and pattern variation in dorsal and lateral surfaces, (A) Karratha, (B) Cape Keraudren, (C) Crab Creek.

intensely surveyed and collected areas in the state (How and Cowan 2006; Doughty *et al.* 2011).

Ctenotus, the most diverse skink genus in Australia, contain many species with extremely localised distributions due to an association with a specific habitat and/ or microhabitat (Wilson and Swan 2013). Ctenotus angusticeps is an excellent example within this category as it is highly specialised in habitat selection. Hence, if the habitat is not 'foraged through' or 'systematically trapped' the species will continue to go undetected. Our targeted surveys employed a simple foraging technique that successfully addressed one of the crucial criteria's outlined for a conservation significant species, which is to 'undertake survey work in suitable habitat and potential habitat to locate any additional populations/occurrences (Threatened Species Scientific Committee 2008).

Since 1990, C. angusticeps has been listed on both state and federal legislation and it has taken 22 vears to address a basic criterion on the mainland. We are also not aware of any research priorities investigating the abundance of this species on Airlie Island in this time. We believe the current listings for many of the 26 'threatened' and 48 'priority' reptile species in Western Australia under the various rankings are, at times ambiguous and can be inconsistent. If they are 'flagged' due to their conservation significance, then the threatening processes for most species should be considered and prioritised measures implemented to address the various criteria. Unfortunately this is not happening due in part to an anthropogenic bias towards avian and mammalian species. During a single survey we greatly expanded the known distribution for C. angusticeps and revealed a unique lifestyle of a lizard inhabiting a very harsh dvnamic environment and subjected to tidal influences and utilising crab holes for shelter. Future studies on how this lizard interacts with its environment will have conservation and management benefits.

In light of this additional information. the conservation status of C. angusticeps requires evaluation. It shows a highly specialised skink inhabiting a naturally fragmented habitat along the northwestern coastline of Western Australia, and it is likely that C. angusticeps will be recorded at other locations between Onslow and Broome (possibly even further afield). However, we recommend that C. angusticeps retains its current 'Vulnerable' status for the following reasons:

- All known populations of this species remain fragmented.
- It is a very highly specialised lizard inhabiting an environment that is harsh and dynamic.
- The natural habitat of this

species is unique and fragmented within the landscape.

- Populations are not known to occur in any protected reserve system on the mainland.
- Relationship between lizards and its environment (e.g. apparent dependency on crab holes for shelter) and their responses to change in the environments is unknown and requires further study.
- The known habitat of this species is prone to development (ports, airports etc) and destruction (4WD vehicles and motorbikes), particularly in a large part of their known range around Karratha, Port Hedland and Broome.
- The habitats of this species are highly vulnerable to both extreme weather (e.g. floods) and potential effects of future climatic change (e.g. rises in the sea level).

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